CHARACTERISATION OF STAPHYLOCOCCUS AUREUS ISOLATED FROM DOOR HANDLES IN THE COLLEGE OF BASIC AND APPLIED SCIENCES, MOUNTAIN TOP UNIVERSITY.

BY

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IN PARTIAL FULFILMENT OF THE REQUIREMENTS FOR THE AWARD OF BACHELOR OF SCIENCE (B.Sc.) IN MICROBIOLOGY

CERTIFICATION

This is to certify that this seminar report titled "CHARACTERISATION OF STAPHYLOCOCCUS AUREUS ISOLATED FROM DOOR HANDLES IN THE COLLEGE AND BASIC AND APPLIED SCIENCES, MOUNTAIN TOP UNIVERSITY" was carried out by SAM-IMONG Favour Ayezele, with matriculation number 18010101041. This project meets the requirements governing the award of Bachelor of Science (B.Sc.) Degree in Microbiology department of biological sciences of Mountain Top University, Ogun State, Nigeria and is approved for its contribution to knowledge and literary presentation.

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DECLARATION

I hereby declare that this seminar report written under the supervision of Dr. Olaleye E.D is a
product of my own research work. Information derived from various sources has been duly
acknowledged in the text and a list of references provided. This seminar report has not been
previously presented anywhere for the award of any degree or certificate.

Sam-Imong Favour Ayezele	
Student	Date

DEDICATION

I would like to dedicate this work to God Almighty, the giver of life and to my family. I am thankful and blessed to have the ultimate support and motivation from my family. My Loving parents Mr. and Mrs. Paul Umoh whose words of encouragement and push for tenacity ring in my ears and my siblings who never left my side.

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TABLE OF CONTENT

Table of Contents Title Page	i
Certification	ii
Declaration	iii
Dedication	iv
Acknowledgement	V
Table of content	V
List of Figures	X
List of Tables	xi
Abstract	xii
CHAPTER ONE	
1.0 INTRODUCTION	1
1.1 Background of the study	2
1.2 Statement of Problem.	2
1.3 Aim and Objectives of the Study	2
1.4 Scope of the Study	2
1.5 Significance of the Study	3
CHAPTER TWO	
2.0 LITERATURE REVIEW	4
2.1 Staphylococcus aureus colonisation and Infection	5
2.2 Virulence factors expressed by <i>Staphylococcus aureus</i>	6
2.3 Mode of transmission	7
2.3.1 Fomites (door handles) as a vector of transmission	7
2.3.2 Human hands as a major transmitter of microorganism to environmental surfaces	8

2.4 Infections caused by <i>Staphylococcus aureus</i>	8
2.4.1 Skin Infections	9
2.4.2 Food Poisoning	10
2.4.3 Bacteremia	11
2.4.4 Toxic Shock Syndrome	11
2.4.5 Septic Arthritis	11
2.5 Immune responses against <i>Staphylococcus aureus</i> Infections	12
2.6 Antibiotic used in the treatment of <i>Staphylococcus aureus</i> Infections	13
2.6.1 Nafcillin	13
2.6.2 Cefazolin	13
2.6.3 Oxacillin	14
2.6.4 Vancomycin	14
2.6.5 Daptomycin	15
2.6.6 Linezolid	15
2.7 Antibiotic resistance in <i>Staphylococcus aureus</i>	15
2.8 Pathogens associated with door handles	16
2.8.Staphylococcus aureus	16
2.8.2 Klebsiella pneumonia	16
2.8.3 Escherichia coli	16
2.8.4 Pseudomonas aeruginosa	
2.9 Prevention and control of <i>Staphylococcus aureus</i> Infections	
2.10 Treatment of <i>Staphylococcus aureus</i> Infections	
CHAPTER THREE	
3.0 MATERIALS AND METHODS	19
3.1 Sample Collection.	

3.2 Materials	19
3.3 Reagents and Equipment used	19
3.4 Media and Reagent	19
3.4.1 FOR Isolation of <i>Staphylococcus aureus</i>	19
3.4.2 For Biochemical Test	19
3.5 Preparation of Culture Media	20
3.5.1 Normal Saline	20
3.5.2 Nutrient Agar	20
3.5.3 Nutrient Broth	20
3.5.4 Mannitol Salt Agar (MSA)	21
3.5.5 Brain Heart Infusion (BHI)	21
3.6 Staphylococcus Species Isolation	22
3.6.1 Primary Enrichment	22
3.6.2 Secondary Enrichment	22
3.6.3 Pure Culture Technique	22
3.6.4 Preservation of <i>Staphylococcus aureus</i> isolates	23
3.6.5 Precautions	23
3.7 Biochemical test for <i>Staphylococccus aureus</i>	23
3.7.1 Gram Staining	23
3.7.2 Catalase Test	24
3.7.3 Coagulase Test	24
3.8 Antimicrobial Susceptibility Testing	24
CHAPTER FOUR	
4.0 RESULTS	25
4.1 Morphological characteristics of the isolates on Mannitol Salt Agar (MSA)	25

4.2 Biochemical characteristics of the isolates	.26
4.3 Prevalence of Staphylococcus aureus isolates	.27
4.4 Antibiotic Susceptibility of the <i>Staphylococcus aureus</i> isolate	27
CHAPTER FIVE	
5.0 DISCUSSION.	31
5.1 Conclusion	32
5.2 References	33

LIST OF FIGURES

Figure		Page
Figure 4.1	Resistance of the isolates to antibiotics	30

LIST OF TABLES

Table		Page
Table 4.1	Morphological characteristics of the isolates on Mannitol salt agar (MSA)	25
Table 4.2	Biochemical characteristics of the isolates	26
Table 4.3	Number of samples and percentage of S. aureus isolates	27
Table 4.4	CLSI Guidelines for the interpretation of zone of inhibition for selected antibiotics in <i>S. aureus</i>	28
Table 4.5	Antibiotic resistance patterns of the isolates	29
Table 4.6	Resistance of the isolates to antibiotics	31

ABSTRACT

Staphylococcus aureus is a commensal organism that resides in skin. Mild to life-threatening diseases can occur if the organism enters into the body especially in an immunocompromised or immunosuppressed individual. This study was carried out in order to determine the prevalence of Staphylococcus aureus on door handles and the antibiotic susceptibility of the organism to some commonly used antibiotics in Mountain Top University. A total of 30 door samples were obtained, 10 each from office doors, lecture rooms and toilets within the College of Basic and Applied Sciences, Mountain Top University. These were cultured and identified using appropriate biochemical tests. A total of 14(46.7%) isolates were identified as Staphylococcus aureus. Toilets doors accounted for the highest prevalence with 64%. The antibiotics susceptibility test revealed that Tetracycline was the most effective of all the test antibiotics against the Staphylococcus aureus isolated. This shows that the door handles harbors Staphylococcus aureus which have the potentials of causing infections. These microorganisms can lead to serious health problems. Therefore, it is necessary to practice good personal hygiene through hand washing and use of hand sanitizer will aid to reduce the incidence of microbial transmission.

KEYWORDS: door handles, antibiotic resistance, staphylococcus aureus.

CHAPTER ONE

1.0 INTRODUCTION

Infectious diseases transmitted via hand contact have been a global health challenge. Gram positive and negative bacteria most especially are found to contaminate contact surfaces e.g; door handles, tables, chairs, windows etc. (Grace et al., 2018). Infectious diseases top the list for causes of death worldwide and contribution to morbidity and mortality cannot be readily quantified due to lack of data for most countries and it remains a global concern (Kones et al., 2017). Infections and/or diseases gotten by contact with environmental surfaces are common cold and sores, conjunctivitis, giardiasis, diarrhea, impetigo, meningitis, pneumonia etc. These diseases are caused by a myriad of bacterial organisms. (Krautkramer et al., 2021). Human hands have been implicated as the major transmitter of microorganisms to environmental surfaces (Tsaku et al., 2017). (Curtis et al., 2003) and (Fewtrell et al., 2007) reported that hands often act as vectors that carry disease-causing pathogens including bacteria and viruses from person to person either through direct contact or indirectly via surfaces. Defective personal hygiene can facilitate the transmission of some of these pathogenic bacteria found in the environment to human hands (Browne et al., 2017). Studies have reported that environmental surfaces which are often touched with hands have higher bacterial load when compared to toilet seats and restroom floor. This outcome might be due to the aggregate contamination of door handles which results from poor sanitary conditions (Fleetwood et al., 2019). Hand washing which is traditional was the first line of defense in preventing the spread of disease; it has been neglected and must be embraced vigorously by families, schools and healthcare professionals. However, many people seem to run water over their hands without using soap and some fail to wash their hands at all after leaving the restroom (Blum et al., 2019). Staphylococcus aureus is important pathogens of human and animals that cause both health care associated infections and community acquired infection (Poolman et al., 2018). It is a pathogen of greater concern because of virulence (Cheung et al., 2021). Its ability to cause a diverse array of life threatening infections and its ability to adapt to different environment condition (Reynolds et al., 2005).

Staphylococcus aureus has been found to be the most frequently isolated pathogen causing blood stream infections, skin and soft tissues infection, bones and joints infection, urinary tract

infection and pneumonia (Klein *et al.*, 2013). Door handles is one of the most implicated probable source of this infections caused by *S. aureus* (Nworie *et al.*, 2012).

1.1 BACKGROUND OF THE STUDY

Staphylococcus aureus is one of the most common microorganisms frequently associated with various diseases, ranging from mild infections of the skin to life-threatening endocarditis, chronic osteomyelitis, pneumonia, and bacteremia (Murray et al., 2005). During the mid-20th century, the introduction and use of antibiotics such as penicillin and methicillin proved successful against S. aureus infections. However, the bacterium quickly acquired resistance to these antibiotics posing an enormous challenge to both veterinary and human health clinicians (Brouillette et al., 2005). Treatment for this bacterium is a concern with the emergence and spread of penicillin-resistant S. aureus. Various antibiotics have proven effective in the treatment of serious staphylococcus aureus infections (McGuinness et al., 2017). Moreover, in the last 20 years, S. aureus clinical isolates with reduced sensitivity to antibiotics and less frequently, with maximum resistance to some antibiotics have emerged (Hidayat et al., 2006).

1.2 STATEMENT OF PROBLEM

Antibiotic resistance has become a major threat to human health worldwide, but its spread through the environment is often overlooked. This study aimed to determine the presence of *Staphylococcus aureus* on doorknobs and their transmission in the environment (Mountain Top University) from human waste, their prevalence and potential for transmission to microbes present in the environment posing a serious threat to public health.

1.3 AIM AND OBJECTIVES OF THE STUDY

This study was therefore designed to determine the presence of *Staphylococcus aureus* on door handles/knobs in the college of Basic and Applied Sciences, Mountain Top University in order to provide scientific information that would have policy relevance, and which will aid the hand washing programmes in Nigeria.

1.4 SCOPE OF THE STUDY

To achieve the objectives, the following procedures were taken

Swabbing of door handles.

- Preparation of culture media.
- Inoculation/streaking of swabs on culture plates.
- Incubation of streaked plates.

1.5 SIGNIFICANCE OF STUDY

This study will provide information about *Staphylococcus aureus* associated with door handles as they pose threat to human health.

CHAPTER TWO

2.0 LITERATURE REVIEW

Microorganisms are found everywhere and are an important part of every ecosystem. In these environments, they are either free-living or parasitic (Sleigh *et al.*, 2018). In some cases, they live as transient contaminants in the fingers or hands, where they pose a major health hazard as a source of hospital-acquired and hospital-acquired infections in the community (Pittet *et al.*, 2016). The increasing incidence of some diseases and their rate of transmission from one community to another have become a major public health concern (Galtelli *et al.*, 2006). Although it is accepted that the risk of infection in the community is generally lower than the risk of infection associated with hospitalized patients, the annual increase in food poisoning cases where outbreaks in the home As the family is a major factor, it is necessary to assess the causes and possible sources (Scott *et al.*, 2010).

Besides daily human interaction, which is a mode of disease transmission, the main source of transmission and spread of community-acquired infections is psoriatic dermatitis (Li *et al.*, 2009). Fomites in frequent contact with humans or the natural habitat of pathogenic organisms are the main source of transmission of infectious diseases (Osterholm *et al.*, 2017). These include door handles for toiletries, showers, toilet seats and faucets, sinks, lockers, tables and chairs, especially those found in public offices, hospitals, hotels, restaurants and toilets (Bright *et al.*, 2010). One of the most likely sources of contamination is doorknobs (Reynolds, 2005). Public offices are full of users who bring their own micro biota and other organisms they have picked up elsewhere and place them on doorknobs/handles for convenient entry and go out (Goldhammer *et al.*, 2006).

However, the risk of termite disease transmission is determined by the frequency of contamination and exposure of the site; the extent of pathogen shedding of the host; probability of transmission of an infectious agent to a susceptible individual; virulence of the organism; immune capacity of the contacts; practice control measures such as disinfectant use and personal hygiene (Reynolds, 2005). Unfortunately, the majority of public toilets found in parks and markets, especially in Nigeria, lack a water system and where they have such systems, water is never available. As a result, it is difficult for users to wash their hands after use, bringing with them contaminants from such utilities (Giannini *et al.*, 2009) and this can lead to outbreaks of *staphylococcus aureus* infections in areas with high prevalence and ongoing cholera outbreaks (Giannini *et al.*, 2009)

2.1 STAPHYLOCOCCUS AUREUS COLONISATION AND INFECTION

Staphylococcus species are Gram-positive, non-motile, non-spore forming microorganisms. S. aureus is the most pathogenic strain of this species with the potential to cause a wide range of diseases both in the community and in hospitals (Otto, 2019). As a symbiotic bacterium, S. aureus mainly resides in the nasal cavity of humans and many animals (Warnke et al., 2014). In addition, S. aureus is also found on the skin, inside the oral cavity, upper respiratory tract, lower urogenital tract and gastrointestinal tract of humans (Fournier et al., 2010). In fact, 25 to 30% of the population is colonized by S. aureus at some point, and about 60% of the population is temporarily colonized by this bacterium (Kang et al., 2017).

As mentioned, approximately 30% of healthy individuals are colonized by *S. aureus* (Burian *et al.*, 2010) through a process that reflects competition between host factors and commensal organisms that are resistant to invasion. Colonization and virulence factors of *S. aureus*, facilitating colonization and possible subsequent infection (Chavez & Decker, 2008). Among the skin's structural properties that help prevent *S. aureus* colonization and infection are its low temperature and acidic ph.

S. aureus is a frequent component of human skin and the nasal microflora. However, it can also cause various skin diseases, sometimes leading to systemic infections (Lai et al., 2010). The ability of S. aureus to penetrate and infect the skin depends on specific mechanisms that destroy

the host's defenses. The existence of these multiple resistance mechanisms clearly demonstrates that CAMP plays an important role in host skin defense against *S. aureus* (Zecconi *et al.*, 2013).

The increasing number of reports of virulent and resistant strains of S. aureus has prompted further investigations into the mechanisms that enable this pathogen to cause infection and to transcend the broad spectrum of preventive measures. antibacterial protection on human skin (Cho *et al.*, 2010). We anticipate that future studies will provide more information on the host and microbial determinants involved in the entry and infection of *S. aureus* on the skin. Targeted drug development around highly conserved bacterial resistance mechanisms against host CAMPs is a promising pharmacological approach in the era of highly virulent and drug resistant strains of *S. aureus* (Mediavilla *et al.*, 2012).

2.2 VIRULENCE FACTORS EXPRESSED BY STAPHYLOCOCCUS AUREUS

S. aureus possesses several virulence factors that enable the pathogen to thrive in different host environments and survive in extreme conditions (Liu et al., 2019). S. aureus creates a plaque on the cell surface and secretes virulence factors such as enterotoxins and hemolysis that enhance its pathogenicity (Mairi et al., 2020). Due to its wide distribution in the human environment, S. aureus is considered as one of the pathogens harmful to humans associated with a number of diseases ranging from mild skin infections to more severe and life-threatening systemic infections such as sepsis (Balaji et al., 2017).

It is known that S. aureus produces many virulence factors, such as hemolysis, leukocidines, proteases, enterotoxins, exfoliated toxins and immunoregulatory factors (Chevalier *et al.*, 2010). The expression of these factors is tightly regulated during growth. The agr system, known as the quorum sensing system, is known to play a central role in the regulation of virulence factors.

To date, regarding virulence factor expression studies, bacterial media, such as Trypticase Soy Broth (TSB), Brain-Heart Infusion (BHI) Broth and Luria-Bertani Broth (LB), has been commonly used to culture *S. aureus* (Deurenberg *et al.*, 2007). However, when *S. aureus* infects the host, the circumstances surrounding the bacterial cell are completely different from those in the environment, with the expression pattern of virulence factors in the host suggested to be entirely different from that in the culture medium. (Deleo *et al.*, 2010)

As a result of in vivo experiments, many factors including cellular immunological factors and nutritional conditions are considered to affect the expression of virulence factors, suggesting that the regulatory mechanism of Virulence factors in living organisms are complex (Sommerville *et al.*, 2009

2.3 MODE OF TRANSMISSION

S. aureus is a common skin organism with about 25% of long-term carriers. Due to its presence on the skin, it can be a major cause of contamination of pharmaceutical products due to improper handling or poor aseptic processes and procedures. It can be transmitted by various means, for example through airborne droplets or aerosols, direct contact with contaminated objects (food, water, inanimate objects) or bites.

2.3.1 FOMITES (DOOR HANDLES) AS A VECTOR OF TRANSMISSION

Fomites play a role in the transmission of *s. aureus*, where inanimate objects are considered reservoirs and potential sources of infection (Akinrotoye *et al.*, 2018).Pathogens can be transmitted either directly by surface-to-mouth contact, or indirectly by infecting hands/fingers, which then transmit the pathogen to the mouth, eyes, ears, nose, or genitals (Plata *et al.*, 2009). Body fluids from infected areas can be a source of pathogen transmission to fomites again (Garci *et al.*, 2011). Several studies have shown that pathogens survive on porous layers for several hours, days or even months depending on the number of deposited cells and other conditions related to the microstructure of the surface formite and environmental conditions (Ghebremendhin *et al.*, 2009).

Staphylococci infections can also be passed from person to person. Because *staphylococci* tough, they can live on items like pillowcases or towels long enough to pass on to the next person who touches them.

Staphylococci can make you sick with an infection. You can also get sick from toxins produced by the bacteria.

Staphylococci can survive:

- Drying
- Extreme Temperature
- Stomach acid

2.3.2 HUMAN HANDS AS A MAJOR TRANSMITTER OF MICROORGANISMS TO ENVIRONMENTAL SURFACES

Human hands are considered to be the main carriers of microorganisms on environmental surfaces. (Curtis et al., 2003) and (Fewtrell, 2007) reported that hands often act as carriers of disease-causing pathogens, including bacteria and viruses, from person to person, through direct or indirect contact with surfaces. Poor personal hygiene can facilitate the transmission of some pathogenic bacteria from one environment to another (Taiwo et al., 2004).

Skin and mucous membranes in general form an effective barrier against infection. However, if these barriers are disrupted (e.g. traumatic skin injury or mucosal damage from a viral infection), *S. aureus* can enter underlying tissues or the bloodstream and cause infection.

People who are immunocompromised or wear invasive medical devices are particularly susceptible to infection

Studies have reported that environmental surfaces that are frequently touched by hands have higher levels of bacteria than toilet seats and toilet floors. This result may be due to the overall contamination of the door handle due to poor sanitary conditions (David & Daum 2010). Hand washing, traditionally, is the first line of defense in preventing the spread of disease; it has been overlooked and should be strongly embraced by families, schools and health professionals. However, many people seem to splash water on their hands without using soap, and some do not wash their hands at all after getting out of the toilet (Okon et al., 2009).

2.4 INFECTIONS CAUSED BY STAPHYLOCOCCUS AUREUS

While *Staphylococcus aureus* is usually harmless on the skin, once it is introduced into the bloodstream, it can cause life threatening illnesses. *Staphylococcus aureus* is a major cause of sepsis when introduced into the body via implants, surgical incisions and injectable medicines. Certain strains of *Staphylococcus aureus* are a particular problem in healthcare as they have developed antibiotic resistance (Li *et al.*, 2018).

Staphylococcus infections can range from minor skin problems to life-threatening illness. Signs and symptoms of staph infections vary widely, depending on the location and severity of the infection.

2.4.1 SKIN INFECTIONS

Skin infections caused by staph bacteria include:

Boils

- The most common type of staph infection is the boil.
- This is a pocket of pus that develops in a hair follicle or oil gland.
- The skin over the infected area usually becomes red and swollen.
- If a boil breaks open, it will probably drain pus.
- Boils occur most often under the arms or around the groin or buttocks.(Yaseen *et al.*, 2018)

Impetigo

- This contagious, often painful rash can be caused by staph bacteria.
- Impetigo usually has large blisters that may ooze fluid and develop a honey-colored crust. (Johnson *et al.*, 2018)

Abscess

- Pocket of infection that forms at the site of injury.
- Usually filled with pus.
- Area surrounding the abscess is usually red, painful and swollen and the skin surrounding the abscess can feel warm to the touch. (Yaseen *et al.*, 2018)

Cellulitis

- An infection of the underlying layers of the skin.
- Usually results from a scrape or cut in the skin which allows bacteria to enter, although no injury may be apparent.
- Cellulitis can occur anywhere in the body, but most often occurs on the legs or arms.
- It causes redness and swelling on the surface of the skin.
- Pain at the site of infection.
- Sores or areas of oozing discharge may develop, too. (Cranendonk *et al.*, 2018)

Staphylococcal scalded skin syndrome

- Toxins produced by the staph bacteria may cause staphylococcal scalded skin syndrome.
- Affecting mostly babies and children, this condition includes a fever, a rash and sometimes blisters.
- When the blisters break, the top layer of skin comes off.
- This leaves a red, raw surface that looks like a burn. (Leung et al., 2018)

2.4.2 FOOD POISONING

• Staphylococcus bacteria are one of the most common causes of food poisoning. The bacteria multiply in food and produce toxins that make you sick. Symptoms come on quickly, usually within hours of eating a contaminated food. Symptoms usually disappear quickly, too, often lasting just half a day. (Roussel *et al.*, 2015)

A *staphylococcus* infection in food usually doesn't cause a fever. Signs and symptoms you can expect with this type of staph infection include:

- Nausea and vomiting
- Diarrhea
- Dehydration
- Low blood pressure

2.4.3 BACTEREMIA

- Also known as a bloodstream infection, bacteremia occurs when staph bacteria enter the bloodstream.
- A fever and low blood pressure are signs of bacteremia.
- The bacteria can travel to locations deep within your body to cause infections that affect:
- ✓ Internal organs, such as your brain (meningitis), heart (endocarditis) or lungs (pneumonia)
- ✓ Bones and muscles
- Surgically implanted devices, such as artificial joints or cardiac pacemakers.(Horino *et al.*, 2020)

2.4.4 TOXIC SHOCK SYNDROME

- This life-threatening condition results from toxins produced by some strains of *staphylococcus aureus*.(Burnham *et al.*, 2015)
- The condition has been linked to certain types of tampons, skin wounds and surgery.
- It usually develops suddenly with:
- ✓ A high fever
- ✓ Nausea and vomiting
- ✓ A rash on your palms and soles that looks like a sunburn
- ✓ Confusion
- ✓ Muscle aches
- ✓ Diarrhea
- Stomach pain

2.4.5 SEPTIC ARTHRITIS

- Septic arthritis is often caused by a staph infection.
- The bacteria often target the knees, shoulders, hips, and fingers or toes. Artificial joints may also be at risk of infection. (Corrado *et al.*, 2020)
- Signs and symptoms may include:
- ✓ Joint swelling
- ✓ Severe pain in the affected joint
- ✓ Fever

2.5 IMMUNE RESPONSES AGAINST STAPHYLOCOCCUS AUREUS INFECTIONS

The immune response against S. aureus involves activation of the innate and adaptive immune systems. As the first line of defense against infection, a rapid innate immune response is activated by pattern recognition pathways that detect nonspecific markers of microbial infection (Adhikari et al., 2012). The main result of this is the activation of phagocytic cells such as macrophages and neutrophils. Neutrophils are recognized as key components of the acute response and are of central importance against S. aureus, as reported by the susceptibility of humans and mice to inherited and acquired neutrophil abnormalities to deep infections. The adaptive immune response is activated later in infection, depends on bacterial antigen presentation by antigen-presenting cells, and is influenced by tissue cytokines produced by the innate response born (Allen et al., 2014). Through T-cell activation and B-cell antibody production, the adaptive immune response targets specific bacterial antigens and can be recalled in subsequent infections to provide "memory" against that particular pathogen (Burbelo et al., 2010). The rate of recurrent S. aureus infections indicates that the adaptive memory response is not completely effective. Understanding the contribution of the adaptive immune response in determining susceptibility to S. aureus can help identify risk factors and treatment strategies, and will be critical to successfully exploiting of vaccine development (Bagnoli et al., 2015).

Immune control of acute *S. aureus* infection is highly dependent on the innate immune system. However, adaptive immunity in the form of B cell and T cell responses may influence this control. Adaptive immunity can influence susceptibility to *S. aureus* infections and is of

particular importance in determining the outcome of chronic persistent infections (Boguniewicz *et al.*, 2011). Finding a protective vaccine will depend on our ability to induce an effective adaptive immune response. Recent studies suggest that the induction of an antibody response alone may not be sufficient and that an appropriate vaccine-induced T-cell response is required for protective immunity (Brauweiler *et al.*, 2014). The potential to induce harmful adaptive immune responses has become evident in animal models and clinical trials of vaccination. This highlights the need to further elucidate the components of an effective immune response, a task complicated by the multiple virulence strategies and infection sites employed by this beetle. Each will require targeting with unique strategies for effective prevention and treatment (Choi *et al.*, 2013).

2.6 ANTIBIOTICS USED IN THE TREATMENT OF STAPHYLOCOCCUS AUREUS INFECTION

2.6.1 NAFCILLIN

A semi-synthetic antibiotic related to penicillin, Naficillin is a narrow-spectrum beta-lactam antibiotic drug. It is a beta-lactamase-resistant penicillin that is indicated for the treatment of Staphylococcal infections caused by strains that are resistant to other penicillins, except those caused by MRSA.

MODE OF ACTION

Nafcillin exerts a bactericidal action against penicillin-susceptible microorganisms during the state of active multiplication in the bacterial cell wall synthesis. It inhibits the biosynthesis of the bacterial cell wall by forming covalent bonds with penicillin-binding proteins that play a critical role in the final transpeptidation process (Sakoulas *et al.*, 2014).

2.6.2 CEFAZOLIN

Cefazolin is a broad-spectrum cephalosporin antibiotic mainly used for the treatment of skin bacterial infections and other moderate to severe bacterial infections in the lung, bone, joint, stomach, blood, heart valve, and urinary tract. It is clinically effective against infections caused by staphylococci and streptococci species of Gram positive bacteria.

MODE OF ACTION

In vitro tests demonstrate that the bactericidal action of cephalosporins results from inhibition of cell wall synthesis. By binding to specific penicillin-binding proteins (PBPs) located inside the bacterial cell wall, it inhibits the third and last stage of bacterial cell wall synthesis. Cell lysis is then mediated by bacterial cell wall autolytic enzymes such as autolysins (Mause *et al.*, 2021).

2.6.3 OXACILLIN

Oxacillin is a penicillin beta-lactam antibiotic used in the treatment of bacterial infections caused by susceptible, usually gram-positive, organisms.

MODE OF ACTION

By binding to specific penicillin-binding proteins (PBPs) located inside the bacterial cell wall, Oxacillin inhibits the third and last stage of bacterial cell wall synthesis. Cell lysis is then mediated by bacterial cell wall autolytic enzymes such as autolysins; it is possible that Oxacillin interferes with an autolysin inhibitor. Oxacillin is stable against hydrolysis by a variety of beta-lactamases, including penicillinases, and cephalosporinases and extended spectrum beta-lactamases (Zhou et al.,2017)

2.6.4 VANCOMYCIN

Vancomycin is a glycopeptide antibiotic used to treat severe but susceptible bacterial infections such as MRSA (methicillin-resistant Staphylococcus aureus) infections. It is often reserved as the "drug of last resort", used only after treatment with other antibiotics has failed. The combination of vancomycin and an aminoglycoside acts synergistically in vitro against many strains of *Staphylococcus aureus*.

MODE OF ACTION

Vancomycin, long considered a "drug of last resort" kills by preventing bacteria from building cell walls. It binds to wall-building fragments called peptides, in particular those that end with two copies of the amino acid D-alanine (D-ala). But bacteria have evolved. In addition, vancomycin alters bacterial-cell-membrane permeability and RNA synthesis. There is no cross-resistance between vancomycin and other antibiotics. Vancomycin is not active in vitro against gram-negative bacilli, mycobacteria, or fungi (Umstatter *et al.*, 2020).

2.6.5 DAPTOMYCIN

Daptomycin is a cyclic lipopeptide antibiotic used to treat complicated skin and skin structure infections by susceptible Gram-positive bacteria and bacteremia due to Staphylococcus aureus.

MODE OF ACTION

Daptomycin binds to the cytoplasmic membrane in a calcium-dependent manner. It causes rapid membrane depolarization and a potassium efflux. It is followed by arrest of DNA, RNA and protein synthesis resulting in bacterial cell death. The rapid cell death does not result in rapid bacterial cell lysis (Gray *et al.*, 2020).

2.6.6 LINEZOLID

Linezolid is a synthetic antibiotic which is used for the treatment of infections caused by aerobic Gram-positive bacteria. Its effects are bacteriostatic against both enterococci and staphylococci and bactericidal against most isolates of streptococci.

MODE OF ACTION

Linezolid exerts its antibacterial effects by interfering with bacterial protein translation. It binds to a site on the bacterial 23S ribosomal RNA of the 50S subunit and prevents the formation of a functional 70S initiation complex, which is essential for bacterial reproduction, thereby preventing bacteria from dividing (Leach *et al.*, 2011).

2.7 ANTIBIOTIC RESISTANCE IN STAPHYLOCOCCUS AUREUS

Staphylococcus aureus can exemplify better than any other human pathogen the adaptive evolution of bacteria in the antibiotic era, as it has demonstrated a unique ability to quickly respond to each new antibiotic with the development of a resistance mechanism, starting with penicillin and methicillin, until the most recent, linezolid and daptomycin.

Resistance mechanisms include;

- ✓ Enzymatic inactivation of the antibiotic (penicillinase and aminoglycoside-modification enzymes).
- ✓ Alteration of the target with decreased affinity for the antibiotic.
- ✓ Trapping of the antibiotic (for vancomycin and possibly daptomycin) and efflux pumps (fluoroquinolones and tetracycline).

Complex genetic arrays (*staphylococca*l chromosomal cassette mec elements or the vanA operon) have been acquired by *S. aureus* through horizontal gene transfer, while resistance to other antibiotics, including some of the most recent ones (e.g., fluoroquinolones, linezolid and daptomycin) have developed through spontaneous mutations and positive selection. Detection of the resistance mechanisms and their genetic basis is an important support to antibiotic susceptibility surveillance in *S. aureus*.

2.8 PATHOGENS ASSOCIATED WITH DOOR HANDLES

2.8.1 STAPHYLOCOCCUS AUREUS

Staphylococcus aureus is a species of Gram-positive spherical bacteria that commonly causes surgical and skin infections, bacteremia (bacteria in the blood) and food poisoning. It's a ubiquitous microorganism, and can be found on the skin of warm blooded animals. 20 different species of the Staphylococcus genus have been recognized

2.8.2 KLEBSIELLA PNEUMONIAE

Klebsiella pneumoniae is a <u>Gram-negative</u>, non-motile, <u>encapsulated</u>, <u>lactose-fermenting</u>, <u>facultative anaerobic</u>, rod-shaped <u>bacterium</u>. It appears as a mucoid lactose fermenter on <u>MacConkey agar</u>. Although found in the normal flora of the mouth, skin, and intestines, it can cause destructive changes to human and animal lungs if aspirated.

2.8.3 ESCHERICHIA COLI

Escherichia coli (E. coli) is a bacterium that normally lives in the intestines of both healthy people and animals. In most cases, this bacterium is harmless. It helps digest the food you eat. However, certain strains of E. coli can cause symptoms including diarrhea, stomach pain and cramps and low-grade fever. Some E. coli infections can be dangerous.

2.8.4 PSEUDOMONAS AERUGINOSA

Pseudomonas is a type of bacteria (germ) that is found commonly in the environment, like in soil and in water. Of the many different types of *Pseudomonas*, the one that most often causes infections in humans is called *Pseudomonas aeruginosa*, which can cause infections in the blood, lungs (pneumonia), or other parts of the body after surgery.

These bacteria are constantly finding new ways to avoid the effects of the antibiotics used to treat the infections they cause. <u>Antibiotic resistance</u> occurs when the germs no longer respond to the antibiotics designed to kill them. If they develop resistance to several types of antibiotics, these germs can become multidrug-resistant.

Others pathogens associated with door handles include;

- Enterobacter spp
- Citrobacter spp
- Proteus spp

2. 9 PREVENTION AND CONTROL OF STAPHYLOCOCCUS AUREUS INFECTIONS

Staphylococcus aureus is associated with healthcare-related infections. Its diagnosis is based on performing microbial detection and identification tests within bacterial colonies. A Staphylococcus aureus infection can be prevented and controlled by:

- Practicing good hand hygiene
- Hygienic conditions for medical procedures and appropriate use of antimicrobial drugs
- Proper environmental control methods, including the regular monitoring of air, water and surfaces
- Stringent cleaning and disinfection of equipment and environments
- In clinical environments, isolating patients when appropriate
- Close monitoring of at-risk patients and populations

2.10 TREATMENT OF STAPHYLOCOCCUS AUREUS INFECTIONS

• Treatment usually includes antibiotics and cleaning the infected area.

- However, some staph infections become unresponsive or resistant to common antibiotics.
- To treat an infection caused by antibiotic-resistant staphylococcus, healthcare providers may need to use antibiotics that can cause more side effects.

CHAPTER THREE

3.0 MATERIALS AND METHOD

3.1 SAMPLE COLLECTION

A total of 30 samples were collected from door handles in the College of Basic and Applied Sciences, Mountain Top University.

The samples were collected using sterile swabs moistened using sterile normal saline by swabbing door handles. The samples were properly labeled using reference numbers so that the identity of the office was anonymous and transported to the laboratory for proper analysis,

3.2 MATERIALS

Materials used include petri-dishes, beakers, swab sticks, conical flasks, measuring cylinders, markers, 70% ethanol, marCartney bottles, Eppendorf tubes, cotton wool, test tubes (with their racks), glass slides.

3.3 REAGENTS AND EQUIPMENT USED

Equipment used: Autoclave, weighing balance, distiller, wash bottles, water bath (set at 50°C and 100°C), incubator (set at 37°C), Bunsen burner, oven, inoculating loop.

3.4 MEDIA AND REAGENT

3.4.1 FOR ISOLATION OF STAPHYLOCOCCUS AUREUS

Nutrient Agar, Nutrient Broth, Mannitol Salt Agar (MSA), Brain Hearth Infusion Broth (BHI), Normal Saline, Distilled water, Ethanol.

3.4.2 FOR BIOCHEMICAL TEST

Crystal violet, Iodine, Alcohol (95%), Safranin, 3% Hydrogen peroxide, blood plasma.

3.5 PREPARATION OF CULTURE MEDIA

3.5.1 NORMAL SALINE

Normal saline is a crystalloid fluid. By definition, it is an aqueous solution of electrolytes and other hydrophilic molecules. It can come in various concentrations; the two specifically addressed are 0.9% and 0.45%.

PREPARATION

0.9g of NaCl was dissolved in 100ml of distilled water based on manufacturer's instructions in a conical flask and was mixed thoroughly. The conical flask is then closed with a cotton wool that is wrapped in aluminum foil. The mixture was heated for a while to dissolve the powder completely and was then sterilized by autoclaving at 121°C for 15mins. 7ml of the mixture was then dispensed into various MarCartney bottles and kept for use.

3.5.2 NUTRIENT AGAR

Nutrient agar was prepared according to the manufacturer's instruction for isolation and detection of total count of mesophilic organism.

PREPARATION

The dehydrated medium was dissolved in the appropriate volume of distilled water i.e. 28g of Nutrient agar in 1 liter of distilled water based on manufacturer's instructions in a conical flask and mixed thoroughly. The conical flask is then closed in cotton wool that is wrapped in aluminum foil. The mixture was heated for a while to dissolve the powder completely and was then sterilized by autoclaving at 121°C for 15minutes. The medium was then allowed to cool to a range of 45-50°C and poured aseptically into sterile petri dishes and left to solidify.

Note: The medium appears opalescent and is light amber in color.

3.5.3 NUTRIENT BROTH

Nutrient broth is the nutrient agar that lack of the solidifying agent, agar powder. They remain in liquid form at room temperature and are usually used to maintain the stocks of microorganisms. In general, they are used to grow fastidious organisms.

PREPARATION

Add 13g of nutrient broth powder in 1L of distilled water. Mix and dissolve them completely. Pour them into the final containers (eg. conical flask). Sterilize by autoclaving at 121°C for 15 minutes.

3.5.4 MANNITOL SALT AGAR (MSA)

Mannitol Salt Agar (MSA) is used as a selective and differential medium for the isolation and identification of *Staphylococcus aureus* from clinical and non-clinical specimens.

PREPARATION

Suspend 111 g of the powder in 1 liter of distilled or deionized water. Mix well. Heat to boil for 1minute shaking frequently until completely dissolved. Sterilize in autoclave at 121°C for 15 minutes.

3.5.5 BRAIN HEART INFUSION (BHI)

Brain Heart Infusion (BHI) broth is a general-purpose liquid medium for the culture and maintenance of a wide range of fastidious and non-fastidious microorganisms, including aerobic and anaerobic bacteria, yeast, and molds from a variety of clinical and non-clinical specimens.

PREPARATION

The dehydrated medium was dissolved in 1 liter of distilled water based on manufacturer's instructions in a conical flask and was mixed thoroughly. The conical flask is then closed with a cotton wool that is wrapped in aluminum foil. The mixture was heated for a while to dissolve the powder completely and was then sterilized by autoclaving at 121°C for 15mins. 5ml of the 0.1% was then dispensed into various test tubes.

3.6 STAPHYLOCOCCUS SPECIES ISOLATION

30 sterile swab sticks were dipped in normal saline to become moist and was used to swab 30 different door handles in the College of Basic and Applied Sciences, Mountain Top University and taken to the laboratory.

3.6.1 PRIMARY ENRICHMENT

In the laboratory 20 swabs sticks were thoroughly mixed to suspend the microorganisms into the buffered normal saline solution. The suspension was inoculated into Mannitol Salt Agar media prepared Mannitol salt agar a using the streaking plate method then incubated at 35°C for 18-24 hours. Mannitol Salt Agar is used or the selective isolation and enumeration of *staphylococcus aureus* from clinical and non-clinical materials. Only *staphylococcus aureus* grow on agar media

containing 7.5% sodium chloride. Addition of 7.5% sodium chloride to phenol red mannitol salt agar results in an improved medium for the isolation of plasma coagulating *staphylococci*. Mannitol fermentation, as indicated by a change in the phenol red indicator, aids in the differentiation of *staphylococcal* species.

The identity of the isolates was confirmed by standard laboratory methods which included gram staining, catalase and coagulase test.

3.6.2 SECONDARY ENRICHMENT

The remaining ten swab sticks with the samples was dipped into 7mls Nutrient broth in marCartney bottles and incubated for 24hrs in 37°C. After incubation for 24hrs. Colonies counted on plate were sub-cultured from Nutrient broth to the newly prepared Mannitol Salt Agar.

3.6.3 PURE CULTURE TECHNIQUE

Sub culturing is done to purify the isolated bacterial colonies from a mixed culture to a new and single culture, the bacterial isolates transferred or sub-cultured were those which were differentiated based on their colony morphology, shape, color, elevation, and other physical characteristics. Colonies differentiated by morphological characteristics are transferred onto fresh petri dishes containing Nutrient agar. From the primary plates (Mannitol Salt Agar), different isolates were sub-cultured aseptically by streaking onto prepared nutrient agar. For 24hours the plates were incubated at 37°C which resulted in pure culture of the isolated organism.

- On MSA agar *s.aureus* is color yellow which indicates the presence of a mannitol fermenter.
- On Nutrient agar *s.aureus* is color yellow

3.6.4 PRESERVATION OF STAPHYLOCOCCUS AUREUS ISOLATES

Two or three colonies of the isolates were diluted into 5ml of Brain heart infusion (BHI) broth each in a test tube and incubated at 37°C for 18- 24 hour. One ml of the organism was put into a sterile Eppendorf tube containing one ml of sterile 20% glycerol (duplicated) which acts as a cryoprotectant and was stored in a freezer at -4 °C.

3.6.5 PRECAUTIONS

- Aseptic techniques were observed at every stage of work.
- Personal protective technique was also observed, such as wearing of covered shoe, nose cover, gloves, lab coat, etc.
- Ensured that the inoculating loop cooled before picking the organism when sub-culturing in order not to kill organism of interest.
- Ensured that the petri-dish was incubated inverted.
- Ensured proper timing, most especially during autoclaving.

3.7 BIOCHEMICAL TEST FOR STAPHYLOCOCCUS AUREUS

3.7.1 GRAM STAINING

The gram stain is fundamental to the phenotypic characterization of bacteria. A smear was made on a glass slide and heat fixed. The crystal violet which is the primary stain was flooded on the fixed culture for 60 seconds; the stain was washed off with water. Iodine solution was added to the smear for 60 seconds and was poured off; then rinsed with water. A few drops of ethyl alcohol (decolourizer) was added and rinsed with water immediately after 5 seconds and finally safranin which is the counter stain was added for 60 seconds and washed off, the the smear was left to air dry. After drying of the slide, it was observed under the microscope. Gram staining was done to find reactions of the bacterial isolates to gram reagents. It helps in classifying and distinguishing bacterial species into two large groups; gram-positive bacteria and gram-negative bacteria.

3.7.2 CATALASE TEST

Catalase test is used in differentiating Staphylococci (which produces catalase enzyme) from Streptococci (which does not produce catalase enzyme). 1Ml of hydrogen peroxide solution is placed in a test tube, and a small amount of bacteria growth was added by wood stick. The formation of air bubbles indicates a postive result.

3.7.3 COAGULASE TEST

Coagulase test is a biochemical test that is used to differentiate Staphylococcus aureus (coagulase positive) from other Staphylococci species (coagulase negative) on the basis of the ability to produce the coagulase enzyme. This enzyme clots the plasma by converting fibrinogen to fibrin. The test was done by placing a drop of plasma on the slide and the isolated organism was added and mixed gently. Within 10 seconds, positivity was detected by the clumping bacterial cells.

3.8 ANTIMICROBIAL SUSCEPTIBILITY TESTING

Antimicrobial susceptibility testing was done by the use of Kirby Bauer disk diffusion method under Clinical Laboratory Standards Institute (CLSI) guidelines. Five colonies of the organisms were emulsified in 5 mls of sterile normal saline and mixed well, the turbidity was compared to 0.5 Mac Farland standard.

A sterile cotton swab was used to inoculate the sample into Mueller-Hinton agar plates and allowed to dry. The following antibiotics were used; oxacillin (3 μg cefoxitin), 20/10μg amoxicillin/clavulanic acid,10μg gentamycin, 30μg ceftazidime, 30μg vancomycin, 5μg Levofloxacin, 10μg ampicillin, 30μg tetracycline, 1.25/23.75μg trimethoprim-sulfamethoxazole and 15μg erythromycin. Zone of inhibitions were determined by measuring the size of clear zones and compared to the CLSI guidelines.

The reporting was done by indicating Resistant, Intermediate or Sensitive.

CHAPTER FOUR

4.0 RESULTS

4.1 MORPHOLOGICAL CHARACTERISTICS OF THE ISOLATES

Following inoculation of a sample onto MSA, one colony with the right morphology characteristic of *S. aureus* was picked and then subcultured onto nutrient agar to obtain a pure culture. The colony selected/picked from the MSA culture of each sample had the following characteristics: have an entire margin, convex elevation, small size, round shape and an opaque transparency (as shown in Table 4.1). Most of the isolates fermented mannitol

Table 4.1 Morphological characteristics of the isolates on Mannitol Salt Agar

Isolate	Margin	Surface	Mannitol fermentation	Elevation	Color (MSA)	Transparency	
S1	Entire	Smooth	NO	Convex	Red	Opaque	
S2	Entire	Smooth	NO	Convex	Red	Opaque	
S3	Entire	Smooth	NO	Convex	Red	Opaque	
S4	Entire	Smooth	YES	Convex	Golden yellow	Opaque	
S5	Entire	Smooth	NO	Convex	Red	Opaque	
S6	Entire	Smooth	NO	Convex	Red	Opaque	
S7	Entire	Smooth	YES	Convex	Golden yellow	Opaque	
S8	Entire	Smooth	NO	Convex	Red	Opaque	
S9	Entire	Smooth	NO	Convex	Red	Opaque	
S10	Entire	Smooth	NO	Convex	Red	Opaque	
S11	Entire	Smooth	YES	Convex	Golden yellow	Opaque	
S12	Entire	Smooth	NO	Convex	Red	Opaque	
S13	Entire	Smooth	NO	Convex	Red	Opaque	
S14	Entire	Smooth	YES	Convex	Golden yellow	Opaque	
S15	Entire	Smooth	NO	Convex	Red	Opaque	
S16	Entire	Smooth	NO	Convex	Red	Opaque	
S17	Entire	Smooth	NO	Convex	Red	Opaque	
S18	Entire	Smooth	YES	Convex	Golden yellow	Opaque	
S19	Entire	Smooth	NO	Convex	Red	Opaque	
S20	Entire	Smooth	NO	Convex	Red	Opaque	
S21	Entire	Smooth	YES	Convex	Golden yellow	Opaque	
S22	Entire	Smooth	YES	Convex	Golden yellow	Opaque	
S23	Entire	Smooth	YES	Convex	Golden yellow	Opaque	
S24	Entire	Smooth	YES	Convex	Golden yellow	Opaque	
S25	Entire	Smooth	YES	Convex	Golden yellow	Opaque	
S26	Entire	Smooth	YES	Convex	Golden yellow	Opaque	
S27	Entire	Smooth	YES	Convex	Golden yellow	Opaque	
S28	Entire	Smooth	YES	Convex	Golden yellow	Opaque	
S29	Entire	Smooth	YES	Convex	Golden yellow	Opaque	
S30	Entire	Smooth	No	Convex	Red	Opaque	

4.2 BIOCHEMICAL CHARACTERISTICS OF THE ISOLATES

Pure cultures of the selected colonies with the right morphology on MSA were then subjected to biochemical tests and results are as shown in *Table 4.2*

Table 4.2 Biochemical characteristics of the isolates

Isolates	Gram Staining	Catalase Test	Coagulase Test	Shape
S1	+	+	-	Rod
S2	+	+	-	Rod
S3	+	+	-	Rod
S4	+	+	+	Cocci
S5	+	-	+	Rod
S6	+	-	+	Rod
S7	+	+	+	Cocci
S8	+	+	-	Rod
S9	+	+	-	Rod
S10	+	+	-	Rod
S11	+	+	+	Cocci
S12	+	-	+	Rod
S13	+	-	+	Rod
S14	+	+	+	Cocci
S15	+	+	-	Rod
S16	+	+	-	Rod
S17	+	+	-	Rod
S18	+	+	+	Cocci
S19	+	-	+	Rod
S20	+	-	+	Rod
S21	+	+	+	Cocci
S22	+	+	+	Cocci
S23	+	+	+	Cocci
S24	+	+	+	Cocci
S25	+	+	+	Cocci
S26	+	+	+	Cocci
S27	+	+	+	Cocci
S28	+	+	+	Cocci
S29	+	+	+	Cocci
S30	+	+	-	Rod

KEY

- Positive is represented as +
- Negative is represented as -

4.3 PREVALENCE OF STAPHYLOCOCCUS AUREUS

A total of 30 samples collected from door handles were processed (10 samples from Offices, 10 samples from lecture rooms and 10 samples from toilets). *Staphylococcus aureus* was isolated from a total of 14 (46.7%) of the 30 door handle swab samples in the College of Basic and Applied Sciences, Mountain Top University.

The results showed that most of the isolates were from the handles of toilet doors; only 2 of the isolates were gotten from office doors (*Table 4.3*)

Table 4.3 Number of samples and percentage of staphylococcus aureus isolates

	Point of Collection	Number of Samples	Number of S. aureus isolates (%)			
1	Offices	10	2 (14%)			
2	Lecture Rooms	10	3 (21%)			
3	Toilet	10	9 (64%)			

4.4 ANTIBIOTIC SUSCEPTIBILITY OF THE STAPHYLOCOCCUS AUREUS ISOLATES

Antibiotic susceptibility testing was performed on all the 14 isolates. Shows the proportion of isolates, classified as susceptible, intermediate or resistant to the antibiotics that were tested and these results are presented in *Table 4.4*. All S. aureus tested showed various level of resistance to Augmentin (50%), Gentamicin (57%), Cefuroxime (50%), Ciprofloxacin (50%), Cotrimoxazole (78%), Erythromycin (50%), Cefotaxime (29%), Tetracycline (75%), Cephalexin (57%) and Meropenem (64%).

Table 4.4 CLSI Guidelines for Interpretation of zone of inhibition for selected antibiotics to Staphylococcus aureus

Antibiotics	Sensitive	Intermediate	Resistant
AUGMENTIN (AUG)	≥22	-	≤21
CEFOTAXIME (CTX)	≥22	-	≤21
CEFUROXIME (CRX)	≥22	-	≤21
CEPHALEXIN (CEX)	≥22	-	≤21
CIPROFLOXACIN (CIP)	≥21	16-20	≤15
COTRIMOXAZOLE (COT)	≥16	11-15	≤10
ERYTHROMYCIN (ERY)	≥23	14-22	≤13
GENTAMICIN (GEN)	≥15	13-14	≤12
MEROPENEM (MEM)	≥22	-	≤21
TETRACYCLINE (TET)	≥16	11-15	≤10
VANCOMYCIN (VAN)	-	-	-

CLSI, 2020

Table 4.5 Antibiotic resistance patterns of the isolates

Isolates	AUG	CEX	CIP	COT	CRX	CTX	ERY	GEN	MEM	TET	Phenotypic Resistance
S4	R	R	S	R	R	S	R	R	R	S	AUG-CEX-TET-CRX-ERY- GEN-MEM
S7	S	R	S	I	R	R	S	R	R	R	CEX-CRX-CTX-MEM-TET
S11	S	S	R	R	S	R	R	S	R	R	CIP-COT-CTX-ERY-MEM- TET
S14	R	R	R	R	S	R	R	S	S	R	AUG-CEX-CIP-COT-CTX- ERY-TET
S18	R	S	R	R	S	S	S	I	S	R	AUG-CIP-COT-TET
S21	R	S	R	R	S	R	S	S	S	R	AUG-CIP-COT-CTX-TET
S22	R	S	R	I	S	S	S	R	S	I	AUG-CIP-GEN
S23	S	R	S	R	R	S	R	R	R	R	CEX-CRX-ERY-GEN-MEM- TET
S24	S	R	S	R	R	S	I	R	R	S	CEX-COT-CRX-GEN-MEM
S25	S	R	R	R	R	S	R	S	S	R	CEX-CIP-COT-CRX-ERY- TET
S26	S	R	R	I	R	S	I	R	S	R	CEX-CIP-CRX-GEN-TET
S27	S	S	S	R	S	S	I	R	S	R	CIP-GEN-TET
S28	R	S	I	R	S	S	R	R	S	R	AUG-COT-ERY-GEN-TET
S29	R	R	I	R	R	S	R	R	S	R	AUG-CEX-COT-CRX-ERY- GEN-TET
Resistance	50	57	50	78	50	29	50	57	64	78	

KEY: S= Sensitive I= Intermediate R= Resistant

%

Vancomycin (VAN) 30 μ g, Cotrimoxazole (COT) 25 μ g. Erythromycin (ERY) 5 μ g, Cefuroxime (CRX) 10 μ g, Gentamicin (GEN) 10 μ g, Ciprofloxacin (CIP) 5 μ g, Ampicillin (AMP) 10 μ g, Cephalexin (CEX) 1.5 μ g, Meropenem (MEM) 10 μ g, Augmentin (AUG) 30 μ g, Tetracycline (TET) 30 μ g.

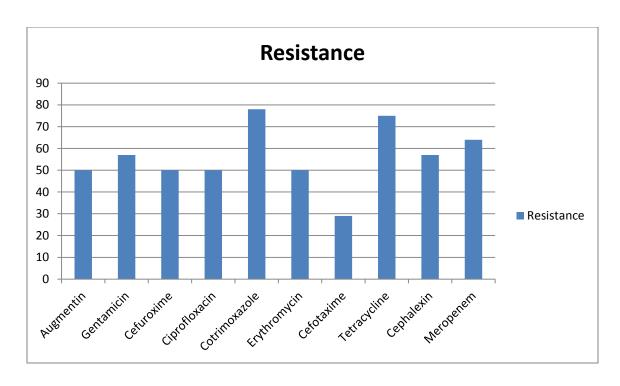


Figure 4.1 Resistance of the isolates to antibiotics

CHAPTER FIVE

5.0 DISCUSSION

This study was designed to isolate and determine the prevalence *S. aureus* from door handles in the College of Basic and Applied Sciences, Mountain Top University and to study its susceptibility to commonly prescribed antibiotics. A total of 14 isolates (46.7%) were obtained and identified as *S. aureus* out of 30 door samples obtained. This prevalence rate is relatively high as compared to the one reported by Owaku *et al* (Owaku *et al.*, 2018) in Nasarawa State University, Keffi Nigeria which found the incidence of *S. aureus* to be 26% out of 100 door samples obtained. In another study, Caroline *et al* (Caroline *et al.*, 2013) reported a higher prevalence of 52% obtained from door handles in selected secondary schools in Nairobi, Kenya which is slightly lower and this may be can be attributed due to the close proximity of the internal parts of the toilet handles to the water closet system and basin sink. Hence contamination is more likely as a result of being exposed to settling air borne microbes from coughing, sneezing, flushing, vector borne spread (flies) and contact with unwashed human hands (Flores *et al.*, 2011). The high frequency of *S.aureus* on toilet door handles can be attributed to the high traffic of students, using these toilets. Offices had the least amount of contamination.

Finally, the higher abundance of *S. aureus* on toilet door handles was also reported by other researchers in their work (Baker *et al.*, 2015).. This could be as a result of high rate of exposure of the door handles to large traffic users who crowd in and out without proper hand hygiene, thereby disseminating their flora to the door handles. Toilet environments usually contain higher microbial loads than other facilities within the University (Alonge *et al.*, 2019)

Environmental factors such as relatively high humidity and moisture content can play crucial role in influencing microbial transfer rates on fomites or hands. It was found that greater microbial carriage and dissemination occurred typically at relative humidity (Emmanuel *et al.*, 2022)

This study shows a high level of resistance to tetracycline and this is because tetracycline is a highly abused drug and overtime many organism have acquired the resistant genes responsible for this antibiotic making them resistant.

5.1 CONCLUSION

This study has revealed that door handles of different places even the most unexpected places are contaminated by a variety of pathogenic and non- pathogenic microorganisms. Hence, door handle surfaces within could therefore act as potential fomites for communicable diseases dissemination. People are encouraged to pay strict attention to personal hygiene practices to avoid the incidence and spread of infections through door handles. This should be considered as a timely warning and proactive measures should be taken to prevent an outbreak of epidemics that could become difficult to handle. These therefore implies that it is possible to be infected with different other microorganisms with greater virulence. Also, the level of multidrug-resistant *S. aureus* isolates is high and should therefore be taken into consideration.

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